Original Article

MRI Cross Sectional Atlas of Normal Canine Cervical Musculoskeletal Structure

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Abstract

Although magnetic resonance imaging (MRI) has been increasingly used as a diagnostic tool for cervical spine injuries in canines, a comprehensive normal MRI anatomy of the canine cervical spine muscles is lacking. Therefore, the purpose of this study was to build a magnetic resonance imaging atlas of the normal cross sectional anatomy of the muscles of the canine cervical spine. MRI scans were performed on a canine cadaver using a combination of T1 and T2-weighted images in the transverse, sagittal and dorsal planes acquired at a slice thickness of 1 mm. Muscle contours were traced manually in each slice, using local osseous structures as reference points for muscle identification. Twenty-two muscles were traced in 401 slices in the cervical region. A three dimensional surface model of all the contoured muscles was created to illustrate the complex geometrical arrangement of canine neck muscles. The cross-sectional area of the muscles was measured at the mid-level of each vertebra. The accuracy of the location of the mapped muscles was verified by comparing the sagittal view of the 3D model of muscles with still photographs obtained from anatomic canine cadaver dissection. We believe this information will provide a unique and valuable resource for veterinary researchers, clinicians and surgeons who wish to evaluate MRI images of the cervical spine. It will also serve as the foundation for ongoing work to develop a computational model of the canine cervical spine in which anatomical information is combined with electromyographic, kinematic and kinetic data.

Keywords: dog; neck; cross sectional anatomy; magnetic resonance imaging

1. Introduction

Biomechanical cervical spine models have been used extensively to evaluate feasibility and potential side effects of surgical procedures and instrumentation as it is currently not feasible to directly measure spinal loading in-vivo (Jaeger et al., 2011). Theoretical and numerical biomechanical models of the human cervical spine have been developed over the last three decades to investigate kinetics and kinematics of the neck (Dugailly et al., 2011). However, these models have not been translated to the canine cervical spine in spite of the high incidence of spinal disorders and injuries (Jeffery et al., 2013). Successful development and implementation of these models in canine spinal studies would require accurate anatomical data of the underlying soft tissues and bone (Sharir et al., 2006). Among the many components which should be incorporated into a model, muscles play a vital role in stability, loading and locomotion as they exert the majority of the required moments to maintain equilibrium in different postures and to perform various tasks (Nussbaum et al., 1995; Vasavada et al., 1998). Studies have shown the substantial effect of muscle forces on cervical spine kinematics and injury potential on the neck structure (Borst et al., 2011). To this extent, comprehensive knowledge of canine muscle properties including estimation of muscle forces and orientation has yet to be established.

The magnitude of the maximum muscle force generation potential in part depends on the muscle morphometric parameters such as physiological cross-sectional area, muscle fiber direction along the length of the muscle, and the muscle attachment site among many other factors (Marras et al., 2001). Therefore, in order to develop an accurate canine specific cervical model, the muscle cross-sectional area (CSA) needs to be directly measured and incorporated.

These geometric properties are usually obtained from anatomic atlases, cadaveric studies or medical images such as computed tomography (CT) and magnetic resonance imaging (MRI). Regardless of technique, regional cross-sectional anatomy is of great importance in identifying the muscle of interest and to determine its biomechanical properties (Zotti et al., 2009). MRI has been used increasingly in dogs as a diagnostic technique for musculoskeletal injuries, joint diseases and soft tissue tumors. It also had has become the preferred imaging modality for investigating articular cartilage, meniscus and ligaments since it provides excellent visualization of soft tissue (Soler et al., 2007; Van Caelenberg et al., 2011; Zook et al., 1989). However a comprehensive search of the literature showed that normal MRI cross-sectional anatomy of the canine neck muscles does not exist. George and Smallwood 1992, had provided an atlas for head and neck using CT in the mesaticephalic dogs. Nevertheless, due to the inability of CT images to differentiate between muscles it is not a comprehensive regional atlas for muscular structure of the canine neck. Hence, the primary aim of this study was to 1) build a comprehensive atlas of cross-sectional anatomy of canine cervical spine muscles using MRI datasets and 2) measure individual CSA of canine cervical spine muscles at each cervical level. This would help to provide a suitable platform for the potential development of a canine specific dynamic biomechanical model of the neck.

We believe that significant insights can be gained from MRI slice base representations. This information will help researchers and clinicians to better evaluate MRI images and enable them to precisely identify and visualize muscular structures of their interest. This project will also be useful for surgeons during pre-operative planning helping identify musculoskeletal structures in the canine neck area. Therefore the purpose of this study was first to provide a cross-sectional anatomy atlas of the canine cervical spine muscles by tracing them with different colors. Second, documenting major force producing neck muscles CSA

2. Materials and methods

2.1 Specimen

A skeletally mature male hound dog (26.0 kg body weight) that was euthanized for reasons unrelated to this study served as the subject. The dog was healthy, with no evidence of joint or spinal disease. It was housed in a single kennel in a room together with other dogs and was fed a standard laboratory dog chow diet with water *ad libitum*. The experimental procedures for this study were reviewed and approved by the local institutional animal care and use committee (IACUC).

2.2 *MRI imaging*

T1 and T2 weighted MRI images were acquired on a 3T MRI scanner (Magnetom Trio, Siemens Healthcare, Erlangen, Germany). Transverse slices of 1 mm thickness were obtained from the skull level and extended caudally to the level of the second thoracic vertebra. MRI examination was performed less than 1 hour after euthanasia to reduce dehydration effects on muscles as much as possible. An MRI-compatible jig was designed to aid in positioning the dog inside the MRI machine. The dog was positioned in ventral recumbency with the thoracic limbs placed in an extended position next to the cervical area and the neck kept in a fairly neutral posture by supporting the neck area with a pillow (Fig.1).

2.3 *Image analysis*

The files generated in DICOM format were retrieved and analyzed with Mimics® software (Materialise NV Technologielaan 15, 3001 Leuven, Belgium). T1-weighted images of all slices from the occiput to the first thoracic vertebra were analyzed. To begin with, bony

structures and muscles were differentiated with the thresholding and region growing applications of the imaging program. Only left sided muscles were traced since it was assumed that spinal musculature would be symmetric. Muscles were traced in each slice based on the visible bony landmarks and the aid of literature about canine anatomy (Boyd et al., 2001; Budras et al., 2007; Kumar, 2012; Miller and Christensen, 1964; Nickel et al., 1992). Each muscle was assigned a separate mask to enhance visualization for outlining of muscle borders and following CSA measurements (Fig 2-8). CSA of the traced muscles were measured at the mid-level of each vertebra (Marras et al., 2001).

2.4 *Validation*

The relative locations of the different neck muscles were compared to photographic images obtained during anatomic canine cadaver dissection. During dissection, the neck muscles were visually identified and separated by removing connective tissues while preserving each muscle's origin and insertion. Following the separation of the muscles, photographs were obtained at different stages of the dissection to compare them with the generated 3D models of the mapped muscles (Fig 9-11).

3. Results

3.1 *Canine cervical muscles mapped from MRI*

Twenty-two canine cervical spine muscles were traced and labeled on 441 transverse MRI image slices (Fig 2-8). Only those muscles that play a role in movement of the neck and partly in the head were considered and grouped as follows: 1. superficial and deep muscle layers of the shoulder girdle; 2. long (superficial, medial, intermediate, deep layers) and short muscles, representing extensors, rotators and neck lateral bending muscles; 3. neck flexors; 4. movers of the head (Nickel et al., 1992; Schomacher and Falla, 2013).

From the superficial shoulder girdle muscle group, the M.trapezius cervicis, M.omotransversarius, M.sternocephalicus, M.cleidomastoideus and M.cleidocervicalis as parts of the M.brachiocephalicus; from the deep shoulder girdle muscle group, the M.rhomboideus and M.serratus ventralis were included. The long neck muscles were represented by the M.splenius as the superficial layer, the M.longissimus (capitis and cervicis), M.longissimus thoracis and M.iliocostalis thoracis as part of the medium layer and the M.spinalis et semispinalis cervicis, M.semispinalis capitis (biventer and complexus) and M.multifidus cervicis as the deep layer. The short neck muscles were represented by the M.intertransversarii cervicis only. On the ventral neck area, the M.longus colli and M.scalenus were traced as the neck flexors. Included muscles that are considered movers of the head were the M.longus capitis, M. rectus capitis dorsalis major, the M.obliquus capitis (caudalis and cranialis) and the M.rectus capitis lateralis and M.rectus capitis ventralis. The M.cleidobrachialis, M.interspinal cervicis, and the M.rectus capitis dorsalis minor were not traced. A three dimensional (3D) model of all the identified and contoured muscles was created to illustrate neck muscle location in 3D (Fig 8-9).

3.2 *Cross-sectional area of canine cervical muscles*

The CSA were measured in all 22 canine cervical muscles that were discriminated in MRI images and results are shown in Table 1. Based on the length of the muscles, in this study, they were grouped in three categories - long, medium and short muscles. The long muscles are defined as extending either over the whole neck area, from C1-C2 into the thoracic area This group includes the M.rhomboideus, M.splenius, M.semispinalis capitis (biventer and complexus), and M.longissimus capitis. Or they are defined as extending over six vertebrae, with additional segmental insertions / origins such as the M.longus capitis, M.longus colli, M.intertransversarii cervicis. Medium muscles are defined as extending over either five

vertebrae such as the M.cleidocervicalis, M.sternocephalicus, M.cleidomastoideus, M.omottransversarius, M.trapezius cervicis, M.spinalis et semispinalis cervicis, M.multifidus cervicis and M.longissimus cervicis or over four vertebrae including the M.serratus ventralis and M.scalenus. Short muscles are defined as those presented at only one level such as the M.obliquus capitis cranialis, M.rectus capitis lateralis and M.rectus capitis ventralis or two levels including M.obliquus capitis caudalis and M.rectus capitis dorsalis major.

4. Discussion

This study is part of an effort to develop a biologically-assisted musculoskeletal canine cervical spine biomechanical model. Biomechanical models can be of great value in identifying potential pathways for neck disorders. They represent a quantitative method to evaluate mechanical effects of surgical techniques and interbody implants on spine. This research provides fundamental information for the initial development of a canine cervical spine model. However, in order to generalize the outcome of this study, more studies will be necessary that involve more specimens. None the less, this study provides a platform for future investigations. This study, for the first time, has implemented a well-developed precise human biomechanical approach to quantify cervical spine muscle CSA (as opposed to cadaveric studies which have several disadvantages).

In the present study we characterized the anatomical trajectory of the majority of the canine cervical muscles with magnetic resonance imaging in a visual way to build an MRI based cross-sectional atlas of the canine cervical spine muscles. Major force producing muscles of the canine cervical spine were identified by measuring the cross-sectional area of individual muscles.

MRI is a noninvasive cross-sectional imaging technique appropriate for diagnostic, research and teaching purposes (Anastasi et al., 2007) with many advantages compared to

other medical imaging techniques (Alsafy, 2008). Soft tissues such as muscles are not readily observed with other radiological modalities in a way that the borders between different muscles can be distinguished. MRI provides excellent detail of clinically relevant anatomy (Soler et al., 2007). Considering MRI spatial resolution, this imaging technique is more sensitive in discriminating different soft tissues, detecting diseases and distinguishing normal and abnormal structures and has been widely used in dogs in musculoskeletal imaging (Adamiak et al., 2011; Agnello et al., 2008; De Bakker et al., 2014; Schaefer and Forrest, 2006) . However, accurate interpretation and identification of CT and MRI images require comprehensive knowledge of the normal planimetric anatomy of the muscles in the region of interest (Rivero et al., 2005).

This study denotes the musculoskeletal cross-sectional anatomy of the canine cervical spine from the occiput to the first thoracic vertebra. Muscles on MRI images were identified and classified with the help of several anatomy books describing the origin, trajectory and insertion of the muscles in text and drawings (Miller and Christensen, 1964; Nickel et al., 1992) together with photographs of cross-sectional reference cuts (Boyd et al., 2001; Kumar, 2012). The anatomic detail of some muscles showed slight discrepancy especially regarding the photographs of the reference cuts, which was probably due to breed differences, as Boyd et al (2001) used a Beagle for his study compared to the hound used in our study. This made the differentiation and identification of muscles sometimes challenging.

Muscles with several portions were treated as a single muscle body regardless of their different divisions as it was challenging to separate muscles into their distinguished bundles. For instance, the M.intertransversarii cervicis anatomically consisting of the M.intertransversarii dorsalis cervicis, the M.intertransversarii intermedii cervicis and the M.intertransversarii ventralis cervicis, was considered as one single muscle body.

The ability to use all three imaging planes (sagittal, dorsal and transverse) at the same time on one screen in the Mimics® software, made it easier to interactively distinguish and mark the individual muscles. The 3D view substantially aided in the identification of muscles in their complex geometrical arrangement as was described in an earlier study (Jaeger et al., 2011).

The main purpose of this investigation was to map the major muscular actuators of cervical motion. The emphasis was on defining the bulk of the muscle mass, since the origins and insertions have been well established before; for this reason, the muscle bundles were not separated into bundles and no attempt was made to map serrations. We mainly focused on muscles that have major contributions to either moving or stabilizing the neck, regardless of their role in shoulder or limb movements. Twenty-two muscles were identified and mapped, the majority of those do play an active role in movement on the neck and head. We also included some muscles of the shoulder girdle that participate in neck movement (M.sternocephalicus, M.brachiocephalicus, M.rhomboideus and M.serratus ventralis). The M.cleidobrachialis part of the M.brachiocephalicus was not mapped as its insertion on the humerus was not in the field of view of the MR images – the same was true for the M.pectoralis (ssuperficialis and profundus). The M.platysma was not mapped because this muscle was very difficult to identify on MR images due to its flat appearance and origin and insertion points mainly emerging out of aponeuroses. We were not able to identify two of the short neck muscles M.interspinal cervicis and the M.rectus capitis dorsalis minor with confidence. These muscle bellies are small and either span a very short distance between adjacent vertebrae or, in case of the M. rectus capitis dorsalis minor, become merged with the M. rectus capitis dorsalis major. Furthermore, although muscles of the deep layer, such as the M.intertransversarii cervicis were mapped, it was challenging and we were not able to trace them precisely.

Several sequences are reported for use in MRI diagnostic imaging. The T1-weighted images used in the present study to identify the individual muscles, have been reported to give good anatomical detail to identify musculoskeletal structures (Agnello et al., 2008; Baeumlin et al., 2010; Soler et al., 2007; Van Caelenberg et al., 2011). However, it was difficult to map smaller muscles (M.interspinal cervicis and M. rectus capitis dorsalis minor). The muscle size, unclear connective tissue borders between those muscles, and the inability to visually separate muscles due to resolution factors of the 3T MRI machine are the factors that contributed to prevent us from mapping those smaller muscles. The small voxel size of a 3T MRI scanner gives a higher resolution. However, it leads to a much lower signal-to-noise ratio which reduces the ability to identify small structures (Sunico et al., 2012). The same study found that imaging the same specimen with a proton density sequence maximizes the distinction of muscular borders compared to T1 or T2 sequences (Sunico et al., 2012).

In general the CSA measurements are not in agreement with the report by Sharir et al. (2006). This conflict potentially might be due to several reasons, most probably as muscle mass might be different between dogs of different breeds and also between individual dogsMuscle morphometric measurements were taken after dissection of the muscle in Shahir et al (2006). Disturbing muscle connections with the surrounding connective tissue may affect its anatomical properties such as its length and width, which might have influenced measurements of the muscle cross section area. Different approaches were taken to present muscle CSA, which increases the possibility of incompatibility between measurements. Sharir et al., (2006) represented the physiological CSA of an individual muscle as a ratio of muscle volume to its effective fascicle length while in the present study we measured actual CSA for each muscle at different levels on MRI images. Therefore, in the study obtained by Sharir et al., (2006), constant cross section throughout the length of the muscles was assumed.

Although this assumption might be valid for small muscles in the neck region, it is not an appropriate representation for fan shaped muscles that have various attachments, as most of the neck muscles present anatomically. These variations within the reported literature highlight the need for quantitative assessments using up to date technological approaches.

The present study has several limitations. Only a single subject was evaluated, due to the nature of this study being exploratory research. The ventral recumbency position of the dog on the MRI table with the thoracic limbs positioned next to the cervical area with flexed shoulder and elbow joints, might have resulted in altered muscle location and orientation in comparison to a neutral standing position, with extended shoulder and elbow joints. By positioning a pillow underneath the neck area, we tried to keep the neck posture as close as possible to a posture in a standing position, however extended shoulder and elbow joints could not be completely replicated. In spite of the excellent capability of MR images in differentiating between muscles, it was still difficult to distinguish all muscles in the region of interest, especially muscles of the deep layer. Therefore, we primarily aimed to identify muscles in the superficial and medium layer of the neck region, as they are the main actuators in stabilizing and moving the neck. With concurrent computed tomography imaging and evaluation of photographic images of cross-sectional frozen cuts of the same individual, it might have been possible to develop more accurate information to identify the muscles of the deep layer on MR images, but this was beyond of the financial possibilities of this study.

While it is clear that there is likely to be significant breed-to-breed variation particularly in muscle mass, we believe that the data presented in this study can be implemented to develop a canine specific cervical biomechanical model as well as to be used as a guide for future medical imaging investigations such as muscle bilateral symmetry assumption.

Conclusions

The data from this work has allowed for the production of the first comprehensive multi-segmental MRI atlas on the cross-sectional anatomy of the canine cervical spine musculature. We anticipate that the 2D and 3D images from this work will be useful to clinicians and researchers working with the canine cervical spine. They will also serve as the foundation of a more expansive project to combine anatomical and EMG data to produce a computational biomechanical model of the canine cervical spine that can be used to study the impact of both pathology and surgical treatment on spinal kinetics and kinematics.

Conflict of interest statement

None declared.

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Figure legends

Fig.1. MRI of the occipital, cervical and cervico-thoracic area in the sagittal plane. Vertical lines indicate the MRI slice corresponding to the presented images (Fig 2-8). The more cranial slice represents section (a) and the more caudal slice represents section (b) of Figures 2-8.

Fig.2. T1-weighted MRI image at (C1). (a) Cranial C1. (b) Mid-vertebral C1. Muscles are listed dorsal to ventral, left to right.

- C1 and C2
- M.cleidocervicalis
- M.rhomboideus
- M.splenius
- M.cleidomastoideus
- M.semispinalis capitis (Biventer)
- M.semispinalis capitis (Complexus)
- M.longissimus capitis
- M.sternocephalicus
- M. rectus capitis dorsalis major
- M.obliquus capitis caudalis
- M.obliquus capitis cranialis
- M.rectus capitis lateralis
- M. rectus capitis ventralis
- M.longus capitis
- M.longus colli

Fig.3. T1-weighted MRI image at (C2). (a) Cranial C2. (b) Mid-vertebral C2. Muscles are listed dorsal to ventral, left to right.

- Wing of Atlas (C1) and C2
- M.cleidocervicalis
- M.rhomboideus
- M.splenius
- M.cleidomastoideus
- M.semispinalis capitis (Biventer)
- M.semispinalis capitis (Complexus)
- M.longissimus capitis
- M.rectus capitis lateralis
- M.omotransversarius
- M.rectus capitis dorsalis major
- M.obliquus capitis caudalis
- M.sternocephalicus
- M.rectus capitis ventralis
- M.intertransversarii cervicis
- M.longus capitis
- M.longus colli

Fig.4. T1-weighted MRI image at (C3). (a) Cranial C3. (b) Mid-vertebral C3. Muscles are listed dorsal to ventral, left to right.

- C3 and articular process of C4
- M.trapezius cervicis
- M.cleidocervicalis
- M.rhomboideus
- M.splenius
- M.serratus ventralis
- M.omotransversarius
- M.cleidomastoideus
- M.semispinalis capitis (Biventer)
- M.semispinalis capitis (Complexus)
- M.longissimus capitis
- M.longissimus cervicis
- M.intertransversarii cervicis
- M.scalenus
- M.longus capitis
- M.sternocephalicus
- Nuchal ligament
- M.spinalis et semispinalis cervicis
- M.multifidus cervicis
- M.longus colli

Fig.5. T1-weighted MRI image at (C4). (a) Cranial C4. (b) Mid-vertebral C4. Muscles are listed dorsal to ventral, left to right.

- C4 (a, b) and tuberculum ventrale of transverse process of C3 (a)
- M.trapezius cervicis
- M.cleidocervicalis
- M.rhomboideus
- M.splenius
- M.serratus ventralis
- M.omotransversarius
- M.cleidomastoideus
- M.semispinalis capitis (Biventer)
- M.semispinalis capitis (Complexus)
- M.longissimus capitis
- M.longissimus cervicis
- M.intertransversarii cervicis
- M.scalenus
- M.longus capitis
- M.sternocephalicus
- Nuchal ligament
- M.spinalis et semispinalis cervicis
- M.multifidus cervicis
- M.longus colli

Fig.6. T1-weighted MRI image at (C5). (a) Cranial C5. (b) Mid-vertebral C5. Muscles are listed dorsal to ventral, Left to Right.

- C4 articular process and C5
- M.trapezius cervicis
- M.rhomboideus
- M.splenius
- M.serratus ventralis
- M.cleidocervicalis
- M.omotransversarius
- M.semispinalis capitis (Biventer)
- M.semispinalis capitis (Complexus)
- M.longissimus capitis
- M.longissimus cervicis
- M.intertransversarii cervicis
- M.scalenus
- M.longus capitis
- M.cleidomastoideus
- M.sternocephalicus
- Nuchal ligament
- M.spinalis et semispinalis cervicis
- M.multifidus cervicis
- M.longus colli

Fig.7. T1-weighted MRI image at (C6). (a) Cranial C6. (b) Mid-vertebral C6. Muscles are listed dorsal to ventral, left to right.

- C6
- Articulatio humeri (a) and Scapula (b)
- M.trapezius cervicis
- M.omotransversarius
- M.rhomboideus
- M.splenius
- M.serratus ventralis
- M.semispinalis capitis (Biventer)
- M.semispinalis capitis (Complexus)
- M.longissimus capitis
- M.longissimus cervicis
- M.longissimus thoracis and M.illiocostalis thoracis
- M.scalenus
- Nuchal ligament
- M.spinalis et semispinalis cervicis
- M.multifidus cervicis
- M.intertransversarii cervicis
- M.longus capitis
- M.longus colli
- M.sternocephalicus

Fig.8. T1-weighted MRI image at Mid-vertebral level (C7). Muscles are listed dorsal to ventral, left to right.

- C7
- Scapula
- M.trapezius cervicis
- M.rhomboideus
- M.splenius
- M.serratus ventralis
- M.semispinalis capitis (Biventer)
- M.semispinalis capitis (Complexus)
- M.longissimus capitis
- M.longissimus cervicis
- M.longissimus thoracis and M.illiocostalis thoracis
- M.intertransversarii cervicis
- Nuchal ligament
- M.spinalis et semispinalis cervicis
- M.multifidus cervicis
- M.longus colli

Fig.9. Sagittal left lateral view of the superficial shoulder girdle muscles (a) 3D image of mapped muscles. (b) Photographic image of the anatomic canine cadaver dissection. 1 - M.cleidocervicalis; 2 – M.trapezius cervicis; 3 –M. sternocephalicus.

Fig.10. Sagittal lateral view from the left of the superficial and deep shoulder girdle muscles and the superficial long neck muscle (a) 3D image of mapped muscles. (b) Photographic image of the anatomic canine cadaver dissection. 1- M.rhomboideus; 2 - M.splenius; 3 - M.serratus ventralis; 4 - M.omotransversarius.